



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF CHEMICAL
SAFETY AND POLLUTION
PREVENTION

April 9, 2013

MEMORANDUM

Subject: Efficacy Review for Liquidator Electronic Ionization System, EPA File Symbol 68250-R; DB Barcode: D409628.

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Applicant: Liquitech, Inc.
421 Eisenhower Lane South
Lombard, IL 60148

Formulation from the Label:

<u>Active Ingredient</u>	<u>% by wt.</u>
Copper (as metallic)	70.00 %
Silver (as metallic)	30.00 %
Total	100.00 %

I. BACKGROUND

The product, Liquidator Electronic Ionization System (EPA File Symbol 68250-R), is a new product. The product is a copper and silver ionization device that releases copper and silver into potable water lines. The applicant requested to register the product to reduce/control *Legionella* species in the treatment of commercial water distribution systems which have previously been treated in accordance with the Safe Drinking Water Act (SDWA) in commercial and industrial buildings. According to the product operation and installation manual, the disinfection action is attributed to the positively charged copper and silver ions which form electrostatic bonds with negatively charged sites on the microorganism cell wall. These electrostatic bonds create stresses which in turn lead to distorted cell wall permeability, reducing normal intake of life sustaining nutrients, while maintaining target levels of copper and silver below EPA allowable levels for drinking water. Studies were conducted at ATS Labs, located at 1285 Corporate Center Drive, Suite 110, Eagan, MN 55121.

This data package identified as D409628 contained one letter from the applicant's representative to EPA (dated December 13, 2012), EPA Form 8570-35 (Data Matrix), four studies (MRID nos. 4901830-01 through 490183-04), and the proposed label (version (4) dated April 9, 2013).

II. USE DIRECTIONS

This product uses an electronic control unit which is connected to the electronic cell that injects positively charged silver and copper ions into the water to be treated. The ions are attracted to the negative sites on the bacteria cell walls. The Controller is a wall mounted, microprocessor-based device capable of controlling output current levels. The Controller applies a direct current across the Flow Cell's electrodes, stimulating the controlled release of ions. The Liquidator Electronic Ionization System is designed to operate on either 100-120 VAC or 220-240 VAC, 50/60 Hz. The Controller incorporates a digital read out which displays current operating parameters and a keypad from which all system programming is performed. The Controller incorporates two fail-safe (energized) dry contact alarms. The alarm circuits will open when an alarm condition is detected or power is lost. The Flow Cell is installed in the recirculation loop and houses the copper/silver electrodes which release ions into the water distribution systems. The Flow Cell is constructed from high temperature, high pressure, schedule 80 CPVC. The Flow Meter detects the amount of hot water consumption. The current output of the Controller is automatically adjusted up or down based on the amount of water flowing through the Flow Meter. The Remote Environmental Management System bi-directional communication collects, logs, and graphs important operational data as well as providing "Alarm" notifications of malfunctions which can be corrected remotely. Directions within the proposed product operation and installation manual provided the following instructions for the use and preparation of the product:

Installation

The Controller should be installed in an indoor, sheltered area away from direct sources of heat, sunlight and moisture. Power should be supplied to the controller using an electrical circuit with sufficient amperage to accommodate the system's peak current draw. The system can be programmed to automatically change output current on different days and time periods. The Controller also automatically adjusts the output voltage from 0 to 100 volts DC to compensate for changes in water conductivity and flow cell electrode condition to maintain consistent copper/silver ion levels. The Controller applies a direct current across the Flow Cell electrodes,

stimulating the controlled release of copper and silver ions into the domestic water distribution system. The Flow Cell features a quick-connect clamp which simplifies Flow Cell removal for inspection and cleaning. The Flow Meter contains a "Closed Loop Proportional Control" which is capable of adjusting itself to produce the precise amount of ionization needed and ensures no under or over ionization.

Copper Testing

Weekly Copper Testing

Once the Liquidator Electronic Ionization System has been fully commissioned, the level of copper in the water at designated sample sites before peak water consumption has begun should have a targeted level of 0.4 PPM copper, resulting in a target level of 40 PPB silver. These levels are optimal for controlling *Legionella*. The actual copper to silver ratio may vary depending on electrode composition, water chemistry, ambient or transient copper in the water supply, and other conditions. To ensure that proper copper levels are being maintained, the water should be tested at least once each week preferably early in the morning before water consumption has begun. A log sheet is provided in the back of the manual track and record test results.

Testing Copper Levels

A Copper Test Kit is supplied with each Liquidator Electronic Ionization System. The kit is designed to measure copper levels between 0 and 5.0 PPM.

Testing Tips:

- Samples should always be collected in a clean glass or polyethylene bottle.
- Samples should be analyzed as soon as possible after collection.
- Discard tubes that are badly scratched.
- Observe the one year shelf live recommendations for the testing reagent.
- Protect the reagent and other test kit components from sunlight, extreme heat, and extreme cold. The entire kit is best stored in a drawer or cabinet at normal room temperature (65°F to 75°F).

Testing

1. Collect a 50 ml sample in the Water Sample Collecting Bottle.
2. Rinse the Colorimeter Tube with sample water.
3. Fill the rinsed Colorimeter Tube to the 10 mL line with the sample water. Cap and wipe dry.
4. Insert the filled Colorimeter Tube into the Colorimeter's light chamber, being sure to align the index line with the arrow on the meter. Close the lid. This tube is the blank zero.
5. Push the "Read" button to turn the meter on. Press the "zero" button and hold it for two seconds until "bLA" is displayed. Release the button to take a zero reading (0 PPM).
6. Remove the Colorimeter Tube and add 5 drops of Copper Reagent
7. Cap the tube and invert to mix. Wipe tube dry.
8. Insert the Colorimeter Tube into the Colorimeter's light chamber, being sure to align the index line with the arrow on the meter. Close the lid.
9. Push the "read" button. Record the results as PPM copper on the log sheet.

III. AGENCY STANDARDS FOR PROPOSED CLAIMS

There are no Agency standards for the proposed claims.

IV. BRIEF DESCRIPTION OF THE DATA

1. MRID 490183-01: "Time Kill Assay for Antimicrobial Agents, for Copper & Silver Ions Produced by a Liquitech Ion Generator. Test Organisms: *Legionella pneumophila* (ATCC 33153)", by Joshua Luedtke. Study conducted at ATS Labs. Study completion date – October 13, 2012. Project Number A14129.

This study was conducted against *Legionella pneumophila* (ATCC 33153). One lot (Lot 11) of the product, Liquidator Electronic Ionization System, was tested using ATS Labs protocol # LQH01091712.CUST.5 (copy provided). A 5 days culture of the test organism was adjusted to a minimum of 3×10^8 CFU/ml. The product was received ready-to-use. Testing was not conducted in the presence of an organic soil load. A 9.5 ml aliquot of each test substance was transferred to a sterile tube for testing procedures. A 0.5 ml aliquot of the standardized inoculum was added to the test substance representing the start of the test exposure. The inoculated test substance was immediately mixed thoroughly using a vortex mixer. Each inoculated and mixed test substance was exposed for the exposure times of 5 hours, 8 hours, and 24 hours at $37 \pm 1^\circ\text{C}$. At each specified exposure time, the sample was mixed and a 0.1 ml aliquot of the inoculated test substance was transferred to 9.9 ml of neutralizer broth (10^0 dilution). Additional ten-fold dilutions were prepared in Butterfield's buffer. Using a standard microbiological spread plate count procedure, 5.0 ml aliquots of the 10^0 dilution and 0.1 ml of the 10^0 - 10^3 dilutions were plated in duplicate on appropriate recovery medium. All subcultures were incubated at 35 - 37°C in 6.0% CO_2 for 4 days (24 hours subcultures) or 5 days (5 hours and 8 hours subcultures). Following incubation, the subcultures were stored at 2 - 8°C for one day prior to visual examination for the presence or absence of growth, and enumerated. Representative subcultures demonstrating growth were stained and/or biochemically assayed to confirm or rule out the presence of the test organism. Log_{10} and percent reduction were determined for each exposure time. Controls included those for purity, sterility, viability, neutralization confirmation, and test population.

2. MRID 490183-02: "Time Kill Assay for Antimicrobial Agents, for Copper & Silver Ions Produced by a Liquitech Ion Generator. Test Organisms: *Legionella pneumophila* (ATCC 33153)", by Joshua Luedtke. Study conducted at ATS Labs. Study completion date – August 27, 2012. Project Number A13898.

This study was conducted against *Legionella pneumophila* (ATCC 33153). One lot (Lot 4) of the product, Liquidator Electronic Ionization System, was tested using ATS Labs protocol # LQH01072512.CUST.1 (copy provided). A 5 days culture of the test organism was adjusted to a minimum of 3×10^8 CFU/ml. The product was received ready-to-use. Testing was not conducted in the presence of an organic soil load. A 9.5 ml aliquot of each test substance was transferred to a sterile tube for testing procedures. A 0.5 ml aliquot of the standardized inoculum was added to the test substance representing the start of the test exposure. The inoculated test substance was immediately mixed thoroughly using a vortex mixer. Each inoculated and mixed test substance was exposed for the exposure times of 5 hours, 8 hours, and 24 hours at $37 \pm 1^\circ\text{C}$. At each specified exposure time, the sample was mixed and a 0.1 ml aliquot of the inoculated test substance was transferred to 9.9 ml of neutralizer broth (10^0 dilution). Additional ten-fold dilutions were prepared in Butterfield's buffer. Using a standard microbiological spread plate count procedure, 5.0 ml aliquots of the 10^0 dilution and 0.1 ml of the 10^0 - 10^3 dilutions were

plated in duplicate on appropriate recovery medium. All subcultures were incubated at 35-37°C in 6.0% CO₂ for 4 days (24 hours subcultures) or 5 days (5 hours and 8 hours subcultures). Following incubation, the subcultures were visually examined for the presence or absence of growth and enumerated. Representative subcultures demonstrating growth were stained and/or biochemically assayed to confirm or rule out the presence of the test organism. Log₁₀ and percent reduction were determined for each exposure time. Controls included those for purity, sterility, viability, neutralization confirmation, and test population.

3. MRID 490183-03: "Time Kill Assay for Antimicrobial Agents, for Copper & Silver Ions Produced by a Liquitech Ion Generator. Test Organisms: *Legionella pneumophila* (ATCC 33153)", by Joshua Luedtke. Study conducted at ATS Labs. Study completion date – August 27, 2012. Project Number A13900.

This study was conducted against *Legionella pneumophila* (ATCC 33153). One lot (Lot 6) of the product, Liquidator Electronic Ionization System, was tested using ATS Labs protocol # LQH01072512.CUST.3 (copy provided). A 5 days culture of the test organism was adjusted to a minimum of 3×10^8 CFU/ml. The product was received ready-to-use. Testing was not conducted in the presence of an organic soil load. A 9.5 ml aliquot of each test substance was transferred to a sterile tube for testing procedures. A 0.5 ml aliquot of the standardized inoculum was added to the test substance representing the start of the test exposure. The inoculated test substance was immediately mixed thoroughly using a vortex mixer. Each inoculated and mixed test substance was exposed for the exposure times of 5 hours, 8 hours, and 24 hours at 37±1°C. At each specified exposure time, the sample was mixed and a 0.1 ml aliquot of the inoculated test substance was transferred to 9.9 ml of neutralizer broth (10⁰ dilution). Additional ten-fold dilutions were prepared in Butterfield's buffer. Using a standard microbiological spread plate count procedure, 5.0 ml aliquots of the 10⁰ dilution and 0.1 ml of the 10⁰-10³ dilutions were plated in duplicate on appropriate recovery medium. All subcultures were incubated at 35-37°C in 6.0% CO₂ for 4 days (24 hours subcultures) or 5 days (5 hours and 8 hours subcultures). Following incubation, the subcultures were visually examined for the presence or absence of growth and enumerated. Representative subcultures demonstrating growth were stained and/or biochemically assayed to confirm or rule out the presence of the test organism. Log₁₀ and percent reduction were determined for each exposure time. Controls included those for purity, sterility, viability, neutralization confirmation, and test population.

4. MRID 490183-04: "Time Kill Assay for Antimicrobial Agents, for Copper & Silver Ions Produced by a Liquitech Ion Generator. Test Organisms: *Legionella pneumophila* (ATCC 33153)", by Joshua Luedtke. Study conducted at ATS Labs. Study completion date – October 23, 2012. Project Number A14128.

This study was conducted against *Legionella pneumophila* (ATCC 33153). One lot (Lot 10) of the product, Liquidator Electronic Ionization System, was tested using ATS Labs protocol # LQH01091712.CUST.4 (copy provided). A 5 days culture of the test organism was adjusted to a minimum of 3×10^8 CFU/ml. The product was received ready-to-use. Testing was not conducted in the presence of an organic soil load. A 9.5 ml aliquot of each test substance was transferred to a sterile tube for testing procedures. A 0.5 ml aliquot of the standardized inoculum was added to the test substance representing the start of the test exposure. The inoculated test substance was immediately mixed thoroughly using a vortex mixer. Each inoculated and mixed test substance was exposed for the exposure times of 5 hours, 8 hours, and 24 hours at 37±1°C. At each specified exposure time, the sample was mixed and a 0.1 ml aliquot of the inoculated test substance was transferred to 9.9 ml of neutralizer broth (10⁰ dilution). Additional ten-fold dilutions were prepared in Butterfield's buffer. Using a standard microbiological spread

plate count procedure, 5.0 ml aliquots of the 10^0 dilution and 0.1 ml of the 10^0 - 10^3 dilutions were plated in duplicate on appropriate recovery medium. All subcultures were incubated at 35-37°C in 6.0% CO₂ for 4 days (24 hours subcultures) or 5 days (5 hours and 8 hours subcultures). Following incubation, the subcultures were stored at 2-8°C for one day prior to visual examination for the presence or absence of growth, and enumerated. Representative subcultures demonstrating growth were stained and/or biochemically assayed to confirm or rule out the presence of the test organism. Log₁₀ and percent reduction were determined for each exposure time. Controls included those for purity, sterility, viability, neutralization confirmation, and test population.

V. RESULTS

MRID #	Test Organism	Exp. Time	CFU/ml Pop. Cont. (Log ₁₀)	CFU/ml of Survivors	Log ₁₀ Survivors	Percent Reduc.	Log ₁₀ Reduc.
490183-01	<i>Legionella pneumophila</i> (Lot 11)	5 hours	5.0×10^6	3.7×10^3	3.57	99.9%	3.13
		8 hours	(6.70)	2.96×10^2	2.47	99.99%	4.23
		24 hours	5.4×10^6 (6.70)	3.4×10^1	1.53	99.999%	5.20
490183-02	<i>Legionella pneumophila</i> (Lot 4)	5 hours	1.56×10^7	1.7×10^2	2.23	>99.99%	4.96
		8 hours	(7.19)	<2	<0.30	>99.9999%	6.89
		24 hours	1.61×10^7 (7.21)	<2	<0.30	>99.9999%	6.91
490183-03	<i>Legionella pneumophila</i> (Lot 6)	5 hours	1.62×10^7	8.4×10^1	1.92	99.999%	5.29
		8 hours	(7.21)	6	0.78	>99.9999%	6.43
		24 hours	1.53×10^7 (7.18)	<2	<0.30	>99.9999%	6.91
490183-04	<i>Legionella pneumophila</i> (Lot 10)	5 hours	3.9×10^6	2.28×10^2	2.36	99.99%	4.23
		8 hours	(6.59)	1×10^1	1.00	>99.999%	5.59
		24 hours	6.7×10^6 (6.83)	<2	<0.30	>99.9999%	6.53

VI. CONCLUSION

1. The submitted efficacy data (MRID nos. 490183-01 through 490183-04) **support** the use of the product, Liquidator Electronic Ionization System, as a bactericide against *Legionella pneumophila* (ATCC 33153) when used in simulated potable water distribution system with 99.9% reduction in 5 hours contact time to 99.9999% in 24 hours contact time.

VII. LABEL

1. The label revisions below are required for Liquidator Electronic Ionization System (EPA File Symbol 68250-R) for use in a potable water distribution system, as effective against *Legionella pneumophila* (ATCC 33153) within 24 hours of exposure.

- Registrant must add the ATCC number to *Legionella pneumophila*.
- Delete the statement "Bacteria are killed rather than suppressed" or revise to read "*Legionella* are killed rather than suppressed."
- Registrant must remove all claims for residual antimicrobial activity from the label since the product is demonstrated effective only when in use. The term "residual" implies activity once the product is no longer being applied (or in operation).
- Delete the words "waterborne pathogens such as" from the statement "Effective against [preventing, controlling] waterborne pathogens such as *Legionella*"